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LABORATORY DIRECTIONS FOR GENERAL BIOLOGY

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LABORATORY DIRECTIONS

FOR

GENERAL BIOLOGY

BY

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PREFACE

This laboratory manual, in its present edition, differs from the previous one only in containing a number of minor changes that have been made throughout the book in order to make it consistent with the new third edition of the author's "Fundamentals of Biology," which it is designed to accompany. It consists of one hundred independent exercises. They are not all of equal length, but none is too long to be completed within a single two-hour laboratory period. An average class, meeting twice a week, should be able to cover most of the exercises in one semester of seventeen weeks' duration. The plan of the manual is such that, to meet local requirements and preferences, the instructor may readily omit any of the exercises he chooses and add others of his own.

As explained in the author's textbook, the present course in general biology is designed primarily to acquaint the student with the broad underlying principles of the subject, emphasis being placed on those aspects of the subject which seem to be of greatest value in contributing to a liberal education. It is not a substitute for more highly specialized courses in botany and zoology, but rather a preparation for them if the student wishes to pursue his studies further. Therefore, in the following pages, details having only a limited application are purposely omitted or subordinated to those of greater importance.

The directions have been made as simple and concise as possible, and little descriptive matter has been introduced. An inclusion here of numerous statements made in the textbook would be superfluous. Students can readily obtain, before coming to the laboratory, whatever previous knowledge of the subject is necessary. Where such information is included in a laboratory manual, students are inclined to pay scant attention to it, reading carefully only statements telling them what to do. The exercises are designed to direct attention to the most significant features of the material being studied, so that the students' time may be most expeditiously and profitably spent. To this end some ques-

tions are asked, especially where questions will stimulate inquiry to a greater extent than descriptive statements. Questions whose answers are perfectly obvious, however, are omitted, and few questions are asked that cannot be directly answered from the material at hand. Questions designed to promote the drawing of general conclusions or to stimulate discussion, for the most part, also are not included in the manual. These can be asked to better advantage by the teacher himself in connection with recitations based on lectures or on readings in the textbook.

Although these exercises, in successively revised form, have been used for a number of years by classes working under the author's direction, and errors have been eliminated as far as possible, it is by no means presumed that corrections or improvements cannot be suggested by others. To those who offer such assistance, the author will be grateful.

A. W. H.

UNIVERSITY OF CALIFORNIA AT LOS ANGELES,
January, 1940.

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LABORATORY DIRECTIONS

FOR

GENERAL BIOLOGY

GENERAL DIRECTIONS

Supplies.—Before beginning the laboratory work the student will provide himself with the following materials: (1) A set of dissecting instruments consisting of a scalpel, forceps, fine-pointed scissors, pair of dissecting needles, safety-razor blade, dropping pipette, and celluloid ruler. (2) A tripod magnifier. (3) Three microscope slides and a dozen cover glasses; also a piece of soft linen cloth for cleaning them. (4) A loose-leaf notebook cover about 9 by 11 inches in size. (5) A supply of smooth, white, unruled drawing paper; linen ledger is the best. Some ruled paper or a cheaper grade of unruled paper of the same size will be needed for written work. (6) A bottle of black waterproof India ink. (7) A medium hard pencil (2*H*), a pen-holder, six drawing pens (*Spencerian No.* 1), an ordinary ink eraser, and a soft pencil eraser.

Laboratory Work.—At the beginning of the course it is essential to realize the importance of laboratory work and to acquire the proper attitude toward it. Lectures and text-books provide only secondhand information, and, although they are indispensable to the proper study of biology, a real knowledge of the subject will come primarily through individual efforts in the laboratory. Work should not be done in a purely mechanical way, as such a procedure will be without permanent value. The significance of each assignment should be recognized and its bearing on the general subject appreciated. Before beginning an exercise, the student should carefully read the directions and then follow them explicitly. Do not rely upon others for assistance, but learn to work independently. Success in the course

will largely be determined by your ability to see things and to interpret them yourself.

Drawings and Notes.—The object of laboratory work in biology is not to make a set of drawings, but to study organisms first hand. Drawings are not an end but a means—a means of representing your own interpretation of the things observed. Inability to represent on paper what one has seen is nearly always an indication of faulty observation, and not of a lack of ability to draw. Elaborate drawings are not required and should not be attempted. There are two prime requisites for successful laboratory work in biology: *Accuracy* and *neatness*. Every line must stand for a structure. Outlines must be continuous and distinct. In nearly all cases shading is unnecessary and should be avoided. Drawings should be large enough to show necessary details. A common tendency of beginners is to make drawings too small. First draw with pencil, and as soon as possible go over the lines with India ink. Do not make crude sketches in the laboratory and finish them outside. All work must be done in the laboratory.

Notes should be taken on features not brought out by the drawings and must always be written up in ink, in the form of complete sentences. Notes should be made when studying the material, not afterward. Answers to all questions should be written out. Under no circumstances must drawings or statements be copied from reference books or from other members of the class. In describing experiments complete facts should be given as to the object of the experiment, the materials used, the procedure followed, the results obtained, and the conclusions drawn.

Headings and Labels.—Each exercise is to be headed to correspond with the topics in the manual. The name of each individual drawing should appear below it, as in a textbook. The details of the drawing, those parts to which attention has been directed (usually printed in italics in the manual), must be neatly labeled by means of horizontal dotted lines to the right of the drawing. Always indicate the scale, as $\times\frac{1}{2}$, $\times 1$, $\times 2$, $\times 250$, etc. In the manual *m* signifies that the object is to be studied under the tripod magnifier or under a binocular dissecting microscope, while *lp* and *hp* refer to the low and high powers of the compound microscope.

Exercise 1

THE COMPOUND MICROSCOPE

In using the compound microscope it is essential that you thoroughly understand its mechanism. Faulty manipulation will not only hinder your work but may result in costly damage. Remove the instrument from its case, but *do nothing with it* until you have carefully studied the following directions. If unable to identify any of the parts mentioned, refer to the wall chart or ask the assistant.

Notice first the *stage* upon which objects are placed for examination. Only transparent objects or minute translucent or opaque objects in a transparent field are examined with the compound microscope. Light is reflected up through or past the object to be examined by the *mirror*, located beneath the stage. Note that it has a flat and a concave side. See that the *mirror bar* is vertical; it must always be kept in this position. The *diaphragm* is a device for regulating the illumination of the object to be examined. It is located on the underside of the stage. Determine how it operates.

Observe the two sets of lenses. The lens at the top of the tube is called the *ocular* or *eyepiece*, and the lower ones the *objectives*. Do not change the position of the objectives at this time, but note that either one may be put in a straight line with the tube. The shorter objective is the *low-power* objective; the longer one is the *high-power* objective. It is very important that you learn to distinguish these before using the microscope, and to know that the high-power objective is always used *after* the object has been examined under the low power.

Study now the focusing mechanisms. One is the *coarse adjustment*, the other the *fine adjustment*. Note the action of the coarse adjustment, but in turning it be extremely careful not to run the tube so far down that the objective touches the stage. Observe that the tube is *raised* as you turn the coarse adjustment *toward* you. Memorize this fact. The fine adjustment should never be turned more than one or two complete revolutions in either direction. The object is first brought into focus by means of the coarse adjustment; focusing for details is then done with the fine adjustment.

Microscopic objects are always mounted in some transparent medium on thin glass *slides*. A smaller glass, the *cover glass*, is always placed over the object. Both slide and cover glass must be very clean. Some of the objects which you will study will be in the form of *permanent mounts* (or *prepared slides*). Others will be mounted by you as *temporary mounts*. Such a mount you will now make.

With your forceps carefully remove a single leaf from a living moss plant, and place it in the center of a clean slide. Add a drop of water with your pipette, and place a clean cover glass over the leaf. Be careful not to have the upper side of the cover glass wet. Now place the mount on the stage of the microscope, having the object on the slide exactly in the center of the opening in the stage. See that the low-power objective is in a straight line with the tube of the microscope. Using the coarse adjustment, move the tube so that the objective is about 6 mm. (¼ inch) above the slide. Now look through the ocular and adjust the flat side of the mirror until you see a clear field of bright light. Consult the wall chart to see how the mirror should be oriented with reference to the source of light. As you look through the microscope, slowly turn the coarse adjustment *toward* you until the object comes into view. If you have trouble, call the assistant. Move the slide around and examine all parts of the leaf, practicing until you can do this without losing sight of the object.

Before proceeding farther, move the slide so that a portion of the leaf is directly in the center of the field of vision. *Without disturbing the focus*, swing the high-power objective into position. Unless your instrument is an old model, you should be able to do this without having the objective come in contact with the slide. (In using an old microscope, the tube must be raised slightly before the high-power objective can be swung into position.) Now use the fine adjustment to focus for details, never turning it more than one or two revolutions in either direction. This is important, as contact of the objective with the slide will break the cover glass (and thus ruin a permanent preparation) and may scratch the expensive lens. Most microscope damage is incurred through a failure to use the high-power objective with extreme care. In focusing with the fine adjustment, observe that the object has depth as well as length and width. No objects are truly flat. It is only by realizing this fact that you will be able

to study with the microscope intelligently. As you examine various objects, always study them in all three dimensions.

In order to obtain satisfactory results from studies with the compound microscope, and to avoid breakage, learn the following rules:

1. Never focus down on an object unless it is so nearly in focus that you can see its outlines.
2. Never use the high-power objective before first bringing the object into focus and centering it under the low power.
3. Use the coarse adjustment only in combination with the low-power objective, and the fine adjustment only with the high-power objective.
4. Do not turn the fine adjustment more than one or two complete revolutions in either direction.
5. Never allow either objective to come in contact with the stage or with a slide.
6. Never use anything but lens paper for cleaning lenses.
7. Do not unscrew or remove any parts of the instrument.
8. Whenever you are not getting good results with your microscope, call for assistance.

PROTOPLASM AND THE CELL

Exercise 2

LIVING PLANT CELLS

Prepare temporary mounts and study (*hp*) living cells from hairs on a squash petiole, from epidermis stripped from an inner scale of an onion bulb, from a young leaf of waterweed (*Elodea*), or from other favorable material. Study the form and arrangement of the cells and their relative size. As you focus up and down with the fine adjustment, note that each cell has thickness as well as length and width. Distinguish carefully between the non-living *cell wall* and the living *protoplasm* that it encloses. If green bodies (*chloroplasts*) are present, observe their form and distribution; their color is due to a pigment called *chlorophyll*. Observe the strands of *cytoplasm* surrounding clear spaces called *vacuoles*. In some cells there is one large central vacuole around which the cytoplasm occurs as a thin layer. If the cytoplasm shows streaming movements, study carefully. The *nucleus* may or may not be visible in the living cell but often can be rendered so by placing the tissue in a weak iodine solution for a few minutes. Note the size and form of the nucleus, and its position within the cell.

Draw a small group of cells of each kind studied, indicating clearly all the features noted. Represent protoplasm by stippling.

Exercise 3

ROOT-TIP CELLS

Examine a prepared slide showing a longitudinal section through a root tip. These cells have been killed, sectioned, and stained by a special method so as to show features not readily seen in living cells. Study (*hp*) the cells that lie just back of the rootcap, noting the *cell wall*, the dense *cytoplasm*, and the relatively large *nucleus*. Which is stained more deeply, the nucleus or the cytoplasm? Observe that the cytoplasm contains many

small *vacuoles* with minute granules between them. Study the structure of the nucleus, noting the *nuclear membrane*, the granules of *chromatin*, and the dense *nucleoli*. Now study cells a little farther back from the rootcap and observe their greater length. What is the character of the cytoplasm? Has the nucleus increased in size?

Draw several cells from both the younger and older parts of the root tip, indicating very accurately the structure of the nucleus and of the cytoplasm.

Exercise 4

ANIMAL CELLS

Examine a preparation of frog or salamander blood and study (*hp*) the large red corpuscles. These represent a simple type of animal cell. A drop of blood has been placed on the slide and the cells killed and stained. Note that there is no cell wall, the cell being bounded by a thin *plasma membrane*. Study the structure of the *nucleus* and *cytoplasm*, noting the *nuclear membrane*, the *chromatin granules* inside the nucleus, and the minute *cytoplasmic granules* outside the nucleus. Are vacuoles present in the cytoplasm? Do you find one or more nucleoli?

Study a bit of the outer skin of a frog or salamander that has been mounted flat on the slide and stained. Note (*hp*) the shape of the cells, the absence of cell walls, and the structure of the nucleus and cytoplasm. Also study sections of salamander liver, of unfertilized starfish or sea urchin eggs, or other favorable material.

Make drawings of all the kinds of animal cells studied, showing clearly their important features.

UNICELLULAR ORGANISMS

Exercise 5

PROTOCOCCUS

Scrape a small quantity of living *Protococcus* from a piece of bark, mount in a drop of water, and examine (*lp*). Note the numerous green bodies, each representing a cell or small group of cells. Study the structure of a single cell (*hp*), noting the *cell wall* and *protoplasm*. The nucleus is not visible. The green coloring matter, *chlorophyll*, is confined to a single large lobed *chloroplast*, which nearly fills the cell. Find cells that have divided into two, three, or four parts. These illustrate reproduction by cell division, a method called *fission*. The cells arising from a single cell are only temporarily associated, soon separating and carrying on an independent existence.

Draw a single cell of *Protococcus* and several groups of cells.

Exercise 6

YEAST

In a bottle of fermenting fruit juice containing yeast, note the numerous bubbles of carbon dioxide coming to the surface. Prepare a temporary mount and examine (*hp*) the yeast cells. How are they arranged? Is a cell wall present? Is chlorophyll present? Note the presence of *buds* and determine how these are formed. The nucleus is not visible; the large clear spaces inside the cells are vacuoles.

Draw several chains of yeast cells showing bud formation.

Exercise 7

BACTERIA

Bacteria are extremely small colorless plants that can be seen only under the higher powers of the microscope. Mount a drop of water containing decaying organic matter and examine under the microscope. A great deal of patience may be required

to find the bacteria. Use the highest magnification only and very little light. Look for minute colorless bodies, some of which may be moving rapidly across the field of vision. Try to distinguish between the different types seen.

Examine (*hp*) prepared slides showing stained bacteria. Distinguish between the spherical, rod-shaped, and curved forms. Observe in each case whether the cells are separate or arranged in colonies, such as chains, plates, etc.

Make drawings of all available types studied.

In order to prove that the decay of organic matter is brought about by bacteria, perform the following experiment. Fill two clean test tubes with cooled and filtered broth made from meat or beans. Leave one of the tubes open. Reboil the broth in the second tube, and immediately plug with sterilized cotton. After several days compare the odor and appearance of the liquid in each tube, and examine (*hp*) for bacteria. Where decay has taken place, how do you account for the presence of bacteria? Did the liquid in the second tube have access to air? What was the effect of the boiling? Under what conditions will food remain unspoiled?

Exercise 8

AMOEBA

Place on a slide a drop of liquid from a culture containing living amoebae and prepare a temporary mount. Examine carefully (*lp*) until you find one, shutting off most of the light. The unicellular body of *Amoeba* is constantly changing its outline as a result of the extension and withdrawal of finger-like lobes of protoplasm called *pseudopodia*. Study carefully (*hp*) the flowing of the protoplasm as the pseudopodia are put out. Observe that, except for an outer clear layer, the protoplasm is distinctly granular. The cell is bounded by a living *plasma membrane*, there being no cell wall. Note the large clear *contractile vacuole*, observing carefully its behavior. Note also the numerous *food vacuoles* scattered throughout the body and the particles of foreign matter, such as grains of sand.

Make a series of sketches at short intervals to show the change in form exhibited by the animal in locomotion, indicating the direction of movement by means of arrows. Also make a large drawing showing all the structural details seen.

Examine a permanent mount of *Amoeba* (*lp*), noting the dense spherical *nucleus* not readily visible in the living animal. All the protoplasm of the cell exclusive of the nucleus constitutes the *cytoplasm;* carefully study its structure.

Add to the large drawing previously made the details seen in the stained preparation.

Exercise 9

PARAMECIUM

Mount a drop of water containing living paramecia and note (*lp*) the size, shape, and movements of the animals. The constant body form is due to the presence of a firm outer membrane called the *pellicle*, but there is no cell wall as in plants. Which is the *anterior* (forward) end, the rounded or the pointed one? The other is the *posterior* end. Study carefully the manner of swimming. The cell is covered with numerous short *cilia*, which beat in unison, propelling the animal through the water. Does *Paramecium* swim in a straight or in a spiral path? In which direction does it rotate on its axis, from left to right or *vice versa*. Observe and state what happens when an animal encounters an obstacle.

Study (*hp*) a paramecium that has come to rest. If the animals are very active, it will be necessary to prepare a new mount, placing a small piece of lens paper on the slide and adding a drop of the culture liquid and a cover glass. Observe the location and behavior of the two *contractile vacuoles*. Close the diaphragm and study the beating of the cilia. Observe the shallow oblique depression in the anterior half of the animal, called the *oral groove*, and note at its lower end the *oral funnel* through which food enters the cell. If possible study the formation of *food vacuoles* in the oral funnel and describe. In which direction do the food vacuoles travel after entering the cell?

Make a large drawing of a single animal, showing all the structural details that have been seen.

Examine (*lp*) a prepared slide of animals that have been stained to show details not readily visible in the living specimens. Note that two nuclei are present, the larger one being the *macronucleus*, and the smaller one the *micronucleus*. What is their position with reference to each other and where are they located in the cell?

Add to the drawing of a paramecium previously made the details seen in the stained preparation.

Study (*lp*) a prepared slide showing *Paramecium* in several stages of *fission*. Note that each of the two nuclei divides into two parts which move to opposite ends of the body, and that then the whole cell becomes constricted transversely.

Draw several stages of fission.

Exercise 10

VORTICELLA

Prepare a temporary mount of material containing living *Vorticella* and examine (*lp*). Observe the bell-shaped body and the long contractile stalk. Study the distribution of the cilia. What is their function? Of what advantage to the animal if the frequent contraction of the stalk and change of position in the water? Study the structure of a single cell (*hp*). Identify the *oral groove*, *food vacuoles*, *contractile vacuole*, and long U-shaped *macronucleus*.

Make a drawing of a single *Vorticella*, showing all the details seen.

Exercise 11

EUGLENA

Prepare a temporary mount of liquid containing living *Euglena* and study (*lp*). Observe the movements of the organisms, their form, and their color. Is there any change of shape? Study (*hp*) the structure of a single cell. Is a cell wall present? Observe at the *anterior* (forward) end the funnel-shaped opening, the *oral funnel*. Close the diaphragm and try to see the *flagellum*, the long hair-like projection at the anterior end of the body. Find the small red *pigment spot* near the base of the flagellum, and also look in this region for the *contractile vacuole*. Observe the numerous green *chloroplasts*. Do you find any food vacuoles? What inference can you draw in regard to the nutrition of *Euglena*? A spherical *nucleus* is present near the center of the cell but is scarcely visible in the living condition. If you find specimens undergoing fission, note that the division of the cell is longitudinal.

Make several outline drawings to show the variety of shapes that an organism may assume. Draw a single *Euglena* on a large scale, showing all the structural details seen.

Exercise 12

COLONIAL ORGANISMS

A *colony* is an aggregation of individuals, intimately associated structurally, each member of which has little or no dependence upon the others. Examine (*lp* and *hp*) colonies of unicellular algae (such as *Gloeocapsa*, *Nostoc*, *Pediastrum*, *Pandorina*, etc.) of flagellates (such as *Uroglena*), and of protozoans (such as *Carchesium*, *Epistylis*, or *Zoothamnium*). Note in each case the number and arrangement of the cells in the colony, the manner in which they are held together, and the degree of organic union between them. It is not necessary to pay attention to the structure of the individual cells.

Make drawings of the forms studied.

THE LOWER PLANT GROUPS

Exercise 13

SIMPLE MULTICELLULAR ALGAE

Examine (*lp* and *hp*) several simple types of multicellular algae (such as *Ulothrix, Spirogyra, Coleochaete, Draparnaldia*, etc.) noting whether the body is plate-like or filamentous and if filamentous whether branched or unbranched. Note the shape of the individual cells. Are all the cells in the body essentially alike, or are some of them different? Explain. The structure of the individual cells need not be studied. What is the difference between a colony of unicellular individuals and a multicellular organism? What is the significance of structural differentiation in multicellular plants and animals?

Make drawings of the forms studied.

Exercise 14

SPORE FORMATION IN ALGAE

Study the formation of *spores* in a simple green alga (such as *Ulothrix* or *Cladophora*). Compare the cells containing spores with the vegetative cells. How do the spores arise? If living material showing swimming spores is available, observe and describe their escape and behavior. Each spore has the capacity of directly producing a new plant.

Make drawings to illustrate spore formation in the alga studied.

Exercise 15

SEXUAL REPRODUCTION IN ALGAE

Study the formation of *gametes* in a simple green alga (such as *Ulothrix* or *Cladophora*) where there is no obvious differentiation between the two kinds. Compare the cells containing gametes with the vegetative cells and with the spores that the same plant produces. The gametes escape from the cell in which they

are formed, swim through the water by means of cilia, come together in pairs, and fuse to form *zygotes*.

Also study sexual reproduction in an alga where there are two distinct kinds of gametes (such as *Oedogonium* or *Vaucheria*). Compare the cells containing *sperms* (small motile gametes) with those producing *eggs* (large passive gametes). How many eggs are produced in a cell? The union of a sperm with an egg gives rise to a *zygote* or *fertilized egg*.

Make drawings to illustrate sexual reproduction in the alga studied.

In the absence of other material, sexual reproduction may be studied in *Spirogyra*. Observe the elongated vegetative cells arranged in the form of an unbranched filament; each cell contains a single nucleus and one or more spiral, band-like chloroplasts. Note the short lateral tubes put out from the cells of two adjacent filaments, the passage of the contents of one to fuse with those of the other, and the resulting formation of zygotes. This alga is peculiar in that both the fusing gametes are large and non-ciliated, although one is active and the other passive.

Draw several stages in the formation of zygotes in *Spirogyra*.

Exercise 16

BREAD MOLD

Examine (*m*) bread mold (*Rhizopus*) growing on moist stale bread. Observe the white filamentous body of the fungus, called the *mycelium*, and the upright branches arising from it. The latter bear globular, terminal *sporangia* containing numerous spores which turn black as they ripen. The spores are liberated into the air. Mount a bit of mycelium on a slide and examine (*lp*). Are cross walls present or absent? Look for branched, root-like *haustoria*, through which nourishment is absorbed from the bread. How do the spores of bread mold differ from those of the algae previously studied?

Make a drawing showing a portion of the mycelium, a haustorium, and several sporangia.

Exercise 17

LIFE HISTORY OF MOSS

Examine (*m*) individual moss plants, noting the very simple stem and leaves. Wash away the soil from the lower end of the

stem and observe (*lp*) the brown absorptive filaments called *rhizoids*. This plant body is called a *gametophyte* because it produces gametes. These are formed in *sexual organs* borne in a cluster at the upper end of the leafy shoot. Both kinds of sexual organs may occur in the same cluster or in different clusters, depending on the species. Examine (*lp*) preparations showing male organs (*antheridia*) and female organs (*archegonia*). Each antheridium produces many *sperms*, each archegonium a single *egg*.

Make a drawing of a leafy shoot (×2 or larger) with its rhizoids below, and indicate the position of the sexual organs. Also draw an antheridium and an archegonium of a moss.

From the fertilized egg there develops a *sporophyte* consisting of a *foot* embedded in the gametophyte, a long slender stalk or *seta*, and a terminal spore-bearing *capsule*. Examine (*m*) moss sporophytes, noting the lid and cap that come off when the spores are shed into the air. Mount some of the spores and note (*hp*) that each consists of a single thick-walled cell. The spore gives rise to a delicate green filament called the *protonema*. Examine moss protonema (*m*) growing on moist soil. Mount a bit of it and examine (*lp*). Is the protonema branched or unbranched? Look for small buds which give rise to erect leafy shoots.

Draw a moss sporophyte arising from the top of a gametophyte (×2 or larger). Also draw a bit of moss protonema.

Exercise 18

LIFE HISTORY OF FERN

Examine whole fern plants, either living or dried, and note the horizontal underground stem giving rise to roots and to large leaves. Observe the numerous *leaflets* into which the blade is divided. On the underside of the leaf observe (*m*) the *sporangia* arranged in groups called *sori*. Each sorus may be covered with a flap-like membrane (*indusium*). Remove some of the sporangia and prepare a temporary mount. Examine (*hp*), noting the *stalk*, the outer layer of *sterile cells*, and the inner mass of thick-walled *spores*. Note the ring of thick-walled cells, constituting the *annulus*, which extends vertically about two-thirds of the way around the sporangium.

Draw a leaflet (×3) showing the arrangement of the sori. Also draw a single sporangium with its spores.

When a fern spore germinates, it produces a small, green, heart-shaped *gametophyte* that lies flat on the ground. Examine one of these (*m* and *lp*), noting on the under surface the numerous absorptive filaments (*rhizoids*) and the two kinds of *sexual organs*. The male organs (*antheridia*) are found among the rhizoids; the female organs (*archegonia*) are located near the notch. Each antheridium produces a number of swimming *sperms*, each archegonium, a single non-motile *egg*. From the fertilized egg a new sporophyte arises.

Draw a fern gametophyte showing the features observed.

Exercise 19

SELAGINELLA

Examine (*m*) plants of *Selaginella*, noting the stem and small leaves. Terminal *cones* are present, each consisting of two kinds of spore-bearing leaves called *sporophylls*. Remove sporophylls from both the base and the apex of the cone and compare (*m*). Mount a sporophyll of each kind and examine (*lp*). How does the number of *megaspores* (large spores) in a *megasporangium* compare with the number of *microspores* (small spores) in a *microsporangium*? Because two kinds of spores are produced, *Selaginella* is said to be *heterosporous*.

Draw a cone and a portion of the leafy stem (×4) and both kinds of sporophylls with their sporangia. Also draw a single megaspore and a microspore to the same scale.

VEGETATIVE ORGANS OF SEED PLANTS

Exercise 20

ROOT TIP

Prepare a temporary mount of a grass seedling[1] that has been growing in water and study (*lp*). Identify the following regions: (1) The *rootcap*, a protective sheath fitting over the end of the rootlet. (2) The dense *growing region* just behind the rootcap. (3) The *root-hair zone*, characterized by the formation of slender one-celled extensions from the epidermis. Note carefully the varying length and abundance of the root hairs. Carefully examine (*hp*) one of the younger root hairs, noting its connection with the root. Observe the thin layer of *cytoplasm* around the large central *vacuole*. Where is the *nucleus*?

Making a drawing of the rootlet showing its general regions. Also make an enlarged view of a single root hair.

Exercise 21

GROWING REGION OF ROOT

Obtain some seedlings of scarlet runner or of Windsor bean in an early stage of germination. Select one with a straight primary root about 25 mm. (1 inch) long. Make a row of equidistant dots with India ink about 2 mm. apart along the entire length of the root and insert it into the neck of a thistle tube, being careful not to rub off the ink. Loosely pack some moist absorbent cotton in the bulb of the thistle tube and stand its lower end in a bottle partly filled with water. As growth proceeds, note which dots become farthest apart. What does this experiment prove in regard to the region of growth?

Make drawings to show the results of your experiment.

[1] Seeds of redtop (*Agrostis alba*) placed in water will furnish excellent material after 5 or 6 days.

Exercise 22

MATURE ROOT

Examine (*lp*) a prepared slide showing cross sections of a mature buttercup root and observe the central *vascular cylinder* surrounded by the *cortex*. The latter, bounded externally by the single-layered *epidermis*, is composed of undifferentiated tissue called *parenchyma*. In the vascular cylinder there are two kinds of conducting tissues: thick-walled *xylem* and thin-walled *phloem*. The xylem tissue is arranged in the form of a star, the phloem tissue occurring in strands between its rays. Distinguish carefully (*hp*) between the xylem and phloem.

Make a diagram of the cross section, indicating the relation of the principal tissues to one another. Then make a detailed drawing of the vascular cylinder on a larger scale.

Exercise 23

TYPICAL SHOOT

A typical shoot consists of a *stem* bearing *leaves* with *buds* or developing *branches* in their axils. Examine such a shoot and identify its main parts. The leaves arise from the stem at definite places called *nodes*, separated by *internodes*. Compare the length of the internodes on the younger and older parts of the stem. How many leaves are borne at each node in the plant you are studying? Does each leaf consist of a *blade* and *petiole* (leafstalk), or is the latter absent? Is the blade simple or divided into separate leaflets? Is there a pair of small appendages (*stipules*) at the base of the leaf?

Where do the buds arise with reference to the leaves? Explain. In addition to the *lateral buds* is there also a *terminal bud* by means of which the main stem increases in length? Cut a bud longitudinally through the middle and examine (*m*), or study (*lp*) a prepared slide and observe the short stem tip with the minute leaves arising from it. Examine an older portion of the shoot and note the *leaf scars*, places from which the leaves have fallen. If bark has formed, look (*m*) for small pores with raised borders. These are *lenticels*.

Make a drawing of a portion of the shoot studied and of a longitudinal section through a vegetative bud. Also draw an older portion of the shoot to show leaf scars and lenticels.

Exercise 24

YOUNG WOODY STEM

Study (*lp*) a prepared slide showing a cross section of a typical woody stem in its first year of growth. Distinguish between the central *pith*, the surrounding *vascular cylinder*, and the peripheral *cortex*. The pith and cortex are composed mainly of parenchyma tissue. The greater portion of the vascular cylinder consists of *xylem* (wood), while just outside the xylem is a narrow zone of *phloem*. Between the xylem and phloem is a layer of actively dividing cells called the *cambium*. Note the radiating *vascular rays* that traverse the vascular cylinder. Of what kind of cells are they composed? The cortex is bounded externally by the single-layered *epidermis;* note (*hp*) the waxy substance forming a *cuticle* on its outer surface. Observe (*hp*) that the cells of the xylem are thick-walled and mostly devoid of protoplasm. Study the cambium and observe that it gives rise to new phloem toward the outside and new xylem toward the center. In the cortex or in the outer portion of the vascular cylinder you will find groups of thick-walled cells representing *mechanical tissue*. Note their distribution and general character.

Make a diagram of the stem studied, showing the relative extent of its main tissues and their relation to one another. Draw in detail a portion of the cambium with its contiguous xylem and phloem.

Exercise 25

OLDER WOODY STEM

Examine (*lp*) a cross section of an older stem of the same kind as studied in the last exercise. What changes, if any, have taken place in the pith, vascular cylinder, and cortex? Count the layers of wood, and in so doing determine the age of the stem. Each season's accumulation of xylem is known as an *annual ring*. Examine (*hp*) the line of contact between two successive rings and state how it is formed. Do all the vascular rays extend from the pith to the cortex? Explain. Observe the formation of *cork tissue* outside the cortex. Is the epidermis still present? Are the cork cells living or dead? Look for *lenticels*, openings in the corky layer.

Make a diagram of the older stem on the same scale as that of the younger stem previously drawn.

Exercise 26

HERBACEOUS STEM

Study (*lp*) a cross section of the stem of a herbaceous dicotyledon (or of a young woody vine) with separate *vascular bundles*. Observe that the vascular cylinder is broken up by wide extensions of the pith. Does the cambium connect the vascular bundles? Do you find vascular rays within the bundles?

Make a diagram of the stem studied.

Exercise 27

STEM OF A MONOCOTYLEDON

Examine (*lp*) a cross section of a young corn stem and note that the conducting tissues are confined to separate *vascular bundles*. How are these arranged? Look (*hp*) at a single bundle and observe that it is made up of xylem and phloem. There is no cambium present and hence no increase in the size of the vascular bundles.

Make a diagram of the stem studied, showing the approximate number, relative size, and arrangement of the vascular bundles.

Exercise 28

LEAF STRUCTURE

Examine (*hp*) a temporary or a prepared slide showing a cross section of a typical leaf and identify its principal tissues. Note on both sides of the leaf the single layer of *epidermal cells* with a waxy deposit on their outer walls forming the *cuticle*. Do these cells contain chloroplasts? Beneath the upper epidermis may be seen one or two layers of elongated *palisade cells*. Below these is the *spongy tissue* made up of loosely arranged cells with large *intercellular spaces*. The palisade and spongy tissue together comprise the *mesophyll* region of the leaf. Do the cells of the mesophyll have chloroplasts? Find a *vein* and note that it consists of xylem and phloem, the former lying above the latter. Small openings may be seen in the lower epidermis; these are *stomata*, to be studied below.

Make a drawing of a portion of the cross section studied, showing all the tissues identified.

With a sharp scalpel strip a piece of epidermis from the lower surface of a leaf and prepare a temporary mount. Note (*lp*) the form of the epidermal cells and the presence of *stomata*. Observe (*hp*) that a stoma consists of a slit bounded by a pair of *guard cells*. Note the presence or absence of chloroplasts in the guard cells.

Draw a stoma as seen in surface view with some of the surrounding cells of the epidermis.

REPRODUCTION IN SEED PLANTS

Exercise 29

THE FLOWER

Study the flowers that have been provided for this exercise, distinguishing between the outer green *calyx*, the conspicuous *corolla*, the *stamens*, and the *pistil*. The calyx is made up of individual *sepals*. What are their form and number? The corolla is composed of *petals*. What are their form, number, and color? Are they separate and distinct from one another, or united to form a tube? Are all the petals alike in form or not? How many stamens are present? To what are they attached? Distinguish between the pollen-bearing portion (the *anther*) and the stalk (the *filament*). Study (*m*) the pistil, the central organ of the flower. Distinguish between the lower bulbous *ovary* and the slender stalk-like *style* arising from it. What is the character of the tip of the style (the *stigma*)? Is the corolla attached near the base of the ovary so that the ovary is inside the corolla, or is it attached to the top of the ovary so that the ovary is below the corolla? Cut an ovary in half transversely and examine (*m*). How many cavities are present? Note the small white *ovules*. Are there few or many? To what are they attached? Is there any evidence that the pistil is *compound*, that is, made up of two or more *carpels* (simple pistils) that have become more or less united?

Write a detailed description of the flower you have studied, including answers to all the above questions. Draw to scale a front view and a side view of a single flower, a stamen, the pistil, and a cross section of the ovary.

Exercise 30

POLLEN GRAIN AND OVULE

Examine (*lp*) a prepared slide showing a cross section of an anther and note the cavities containing *pollen grains*. Examine (*hp*) a single pollen grain and identify the *tube nucleus* and

generative nucleus. Note the rather heavy *cell wall.* Mount pollen grains[1] which have been growing in a 10 per cent solution of cane sugar and observe (*lp*) the development of the *pollen tubes.*

Make a drawing of a section of a pollen grain showing its internal structure. Also draw a germinating pollen grain.

Obtain a prepared slide of a cross section through the ovary of a lily, or of some other favorable form, and note (*lp*) the position of the *ovules.* Find an ovule that has been cut through the middle and note (*hp*) the large *embryo sac* surrounded by one or two *integuments.* Count the number of nuclei in the embryo sac, if it is not mature there will be less than eight.

Make a drawing of a longitudinal section of an ovule with its embryo sac.

Exercise 31

THE FRUIT

Examine a fruit of some simple type, such as a pod or a berry, if possible in various stages of development. What part or parts of the flower have entered into the formation of the fruit you are studying? Make a cross section or carefully split open the fruit and note the attachment of the seeds. From what have they developed? Is the fruit you are studying dry or fleshy at maturity? If dry does it remain closed or is there some way in which the seeds are liberated?

Draw several stages in the development of the fruit studied. Also draw a cross section or a view showing the fruit split open to show the seeds.

Exercise 32

THE SEED

Study a soaked bean that has been split open and identify the following parts: the outer covering (*testa*); the "halves" of the seed (*cotyledons*); the short stem between the cotyledons (*hypocotyl*); the minute bud at the upper end of the hypocotyl (*plumule*). Note that the cotyledons are large and contain stored food. The hypocotyl, cotyledons, and plumule constitute the *embryo.*

[1] Pollen of nasturtium, sweet pea, or lily germinates promptly.

Make a drawing of a bean seed split open to show the parts of the embryo (×2).

Obtain a soaked grain of corn, cut it in half lengthwise at right angles to its broad face, place a drop of weak iodine solution on the cut surface, and examine (*m*). The *embryo* is stained yellow, the starchy portion of the *endosperm*, dark purple. The embryo has a single *cotyledon*, which is in close contact with the endosperm. Identify the *hypocotyl* with the *plumule* at its upper end. Note the attachment between the hypocotyl and the cotyledon. Where in the corn grain is most of the food stored?

Make a drawing of the corn grain as seen in longitudinal section (×5).

Exercise 33

THE SEEDLING

Examine bean seedlings in various stages of development. Note that the root system has arisen from the lower end of the hypocotyl. Does the hypocotyl continue to grow after the root is formed, or does it remain short? Do the cotyledons appear above the surface of the ground, or do they remain inside the testa beneath the surface? Into what does the plumule develop? What finally happens to the cotyledons?

Make a series of drawings illustrating the development of the bean seedling and write an account of the same, answering the above questions.

Study several stages in the development of the corn seedling, noting the ways in which it differs from that of the bean. What is the function of the single cotyledon? What is the function of the endosperm? Does the hypocotyl continue to grow after the root system has arisen? Does the plumule develop relatively earlier or later than in the bean seedling?

Make several drawings to illustrate the development of the corn seedling and briefly describe.

Exercise 34

REPRODUCTION IN THE PINE

Study staminate and young carpellate cones of any species of pine. Note that they are composed of a number of overlapping

scales. Remove several scales from a staminate cone and examine (*m*). On which side of the scale are the two pollen sacs (microsporangia)? Examine several scales removed from a carpellate cone. Where are the two ovules (megasporangia) situated? Obtain a mature carpellate cone of a pine and note the woody character of the scales. Where are the seeds borne? To what structures in a flower do the cone scales of a pine correspond? To what structures in a cone of *Selaginella* do they correspond? (See Exercise 19.)

Draw a staminate and a young carpellate pine cone (×2 or larger). Also draw individual scales removed from each as seen from two different aspects.

Exercise 35

VEGETATIVE PROPAGATION

Examine plants showing reproduction by means of vegetative organs, such as runners, rhizomes, bulbs, tubers, etc. Also examine cuttings growing in water or in soil, noting the place where the new roots arise.

Make drawings to illustrate vegetative propagation.

METABOLISM AND IRRITABILITY IN PLANTS

Exercise 36

DIFFUSION

Place one or more crystals of some colored salt, such as copper sulphate or potassium bichromate, in a test tube of water and do not disturb for several days. Minute particles of the salt pass out into the water to mingle with the water molecules. At the end of 2 hours measure the height reached by the salt, and measure again in a day or two. In terms of concentration of salt particles per unit volume of water, in what direction has the diffusion of the salt taken place?

Exercise 37

OSMOSIS

Fill a thistle tube with a strong solution of cane sugar or of sodium chloride and firmly tie a piece of parchment (or some other suitable membrane) over the large end. Place in a jar of distilled water so that the membrane is well below the surface, and support with a ring stand and clamp. Attach a long piece of glass tubing to the upper end of the thistle tube. Note the level of the solution and measure any change after 24 and 48 hours. The membrane is freely permeable to the water particles but almost completely checks the free diffusion of the salt or sugar particles through it. In terms of concentration of the dissolved substance, in which direction has the water moved? What force is supporting the liquid in the tube?

Make a drawing of the apparatus and fully explain the results of the experiment.

Exercise 38

PLASMOLYSIS

Place a young leaf of waterweed (*Elodea*), epidermis stripped from a leaf of wandering Jew (*Zebrina*), or other favorable

material in a weak solution[1] of sodium chloride for 2 or 3 minutes and then examine (*hp*), using a drop of the salt solution as a mounting medium. Observe and record by means of three or four drawings successive changes in the appearance of the cells. Then transfer to pure water for a few minutes and draw again. Fully explain the behavior seen.

Exercise 39

ASCENT OF WATER

Cut off a shoot from a vigorous herbaceous plant[2] and immediately immerse the lower end in water colored with eosin or some other dye. Note the rise of the solution in the shoot until it has reached the ends of the veins. Cut thin cross sections of the stem and note (*lp*) the presence of the stain in the vascular system. Through which cells does the water ascend? What has caused a concentration of dye in the leaves?

Briefly describe the experiment and make a diagram of the cross section.

Exercise 40

DEMONSTRATION OF TRANSPIRATION

Insert a shoot of a potted plant[3] into a large Erlenmeyer flask and fasten with a split cork. Water the soil well and in a short time observe the condensation of water vapor on the inside of the flask. Draw the apparatus and fully explain the cause of the appearance of the water drops.

Exercise 41

NECESSITY OF LIGHT IN PHOTOSYNTHESIS

Place a potted plant, having fairly large thin leaves, in the dark for 24 hours. Then fasten a light-screen on one of the younger leaves and place in a well-illuminated situation for several hours. Extract the chlorophyll from the leaf by placing it in alcohol and warming on a water bath for ½ hour. After

[1] Not over 5 per cent strength.

[2] A plant with a pale stem, such as jewelweed (*Impatiens*), is excellent for this exercise.

[3] A sunflower, tomato, or geranium will answer the purpose well.

the leaf is entirely bleached, wash in water and place in a solution of iodine. Observe which parts become dark colored and thus show the presence of starch. Note that no starch is formed in the shaded portion of the leaf. What does this experiment prove?

Write up your results and make a sketch of the leaf.

Exercise 42

RELEASE OF OXYGEN IN PHOTOSYNTHESIS

Place two or three shoots of waterweed (*Elodea*) in a beaker of water. Fill a test tube with water and, holding your thumb over its open end, invert the tube and lower it into the beaker. Then push the cut ends of the shoots a short distance into the tube and place the apparatus in bright sunlight. The gas bubbles that arise from the cut ends of the shoots will gradually displace the water in the test tube. Count the number of bubbles arising per minute from one of the plants; then shade the apparatus and after several minutes count again. After the test tube has become nearly filled with the gas, remove the plants, cork the tube, and take it out of the water. Prepare a glowing splinter of wood, plunge it into the tube, and note its bursting into flame for an instant, thus proving the evolved gas to be oxygen.

Make a drawing of the apparatus and describe the results of your experiment.

Exercise 43

NECESSITY OF OXYGEN IN GROWTH

Place a dozen young oat seedlings in one end of a large U tube with the roots toward the opening. Insert a loose plug of wet absorbent cotton, push it about 1 inch into the tube, and close the opening with a cork. Similarly prepare another U tube. Place the open end of one of the tubes in a jar of water and support with a stand and clamp. Into the open end of the other U tube place a little dry pyrogallic acid and inset a dry cotton plug. Now place this end of the tube in a jar containing a 10 per cent solution of potassium hydroxide, and place the apparatus in a warm, dark situation. Within an hour or two observe that the black solution (potassium pyrogallate) has risen in the tube, having absorbed the oxygen present there. Approximately

what proportion of the air originally present in the U tube has been absorbed? After several days compare the growth of the seedlings in the two tubes. Why has the level of the water in the one tube remained approximately constant? Why was it necessary to keep the seedlings in darkness?

Draw the apparatus; state and explain the results of the experiment.

Exercise 44

RELEASE OF CARBON DIOXIDE IN RESPIRATION

Follow the same method as in the last exercise, but omit the pyrogallic acid. The solution of potassium hydroxide will leave the oxygen for use by the seedlings in respiration, but will absorb any carbon dioxide as fast as it is given off. After several days observe the height of the liquid in both tubes, and explain.

Exercise 45

RESPONSE TO GRAVITY

Obtain two pieces of glass about 15 cm. (6 inches) square, two or three pieces of blotting paper the same size, and some seeds of radish or mustard. Moisten the blotting paper and place nine seeds on it, spacing them evenly. Place the blotters between the two glass plates and tie together with a string. Stand the plates upright in a shallow dish of water and keep in a warm, moist situation. After the seeds have sprouted, notice the growth direction taken by the roots. Then invert the plates so that the roots are pointing in the opposite direction, and after several days observe what changes have taken place. To what stimulus have the roots responded?

State what was done and illustrate your results with a drawing of one of the seedlings before and after the growth direction was changed. Why had the influence of moisture and of light nothing to do with the results in this experiment?

Draw a potted seedling of sunflower or of scarlet runner bean which has been placed in a horizontal position in a dark place for a day or two. Explain the growth direction of the stem tip and the orientation of the leaves. Why was the seedling kept in the dark?

Exercise 46

RESPONSE TO LIGHT

Examine and sketch plants that have been exposed to one-sided illumination for several days. Explain the growth direction of the stem tip and the orientation of the leaves. Of what advantage is this reaction to the plant? If a clinostat is available, place on it a potted plant that has grown in a situation with good overhead illumination, place the plant where the light will strike it largely from one side, and set the clinostat in motion. Explain the results obtained.

THE LOWER ANIMAL GROUPS[1]

Exercise 47

SPONGES

Examine (*m*) preserved specimens of *Grantia* or some similar type of simple sponge. Note the large terminal opening (the *osculum*) at the unattached end and the body wall with its many minute pores. Study a specimen cut lengthwise in half and note the large central cavity (the *cloaca*) and the canal system. The body wall is folded in such a way that the canals that open into the cloaca (the *radial canals*) alternate with those opening to the outside (the *incurrent canals*). The body wall of *Grantia* is supported by calcareous *spicules*. Examine a prepared slide showing isolated spicules (*lp*) and distinguish between the two kinds that are present. Observe a colonial sponge. Here a number of individuals have grown together. Can you determine the limits of a single individual?

Make a drawing of an external view of *Grantia* (×5) and an internal view of one half (×5). Draw the two varieties of spicules.

Exercise 48

HYDRA

Place a living hydra in a watch glass with water and study (*m*) its general form and movements. Observe the cylindrical *stalk* with its finger-like *tentacles*. How many tentacles does your specimen have? Note that the tentacles surround a conical elevation, the *hypostome*, in the center of which is the *mouth*. At the lower end of the stalk is the *basal disk*, which becomes attached to objects in the water. Gently touch the animal and describe its reaction.

[1] Representatives of the lowest animal group, the protozoans, have already been studied, and no more attention will be given to it here (see Exercises 8 to 11).

Now transfer the specimen to a slide and make a temporary mount, using bits of broken glass under the cover to prevent crushing. Use very little light. Study the structure of the tentacles (*lp*), noting the small, oval, tubercular bodies occurring in ridges; these are *stinging cells.* Note that both stalk and tentacles are hollow. Look for *buds* on the stalk of your specimen, lateral outgrowths that become detached and give rise to new individuals. Sexual organs may be present on the stalk, appearing as small swellings. The male organs, called *testes*, are located just below the hypostome, while the female organs, called *ovaries*, may be found about one-third the length of the stalk above the basal disk. Both male and female organs may occur on the same or on different individuals, depending on the species.

Make a drawing of a hydra in both contracted and expanded conditions (*m*). Also draw a portion of a tentacle on a larger scale to show external details of structure. If specimens with buds or with sexual organs were seen, draw.

Examine a prepared slide showing a cross section through the stalk, and note the *body wall* surrounding the central cavity, the *coelenteron.* The latter is continuous into the tentacles. Observe (*hp*) that the body wall is composed of two layers of cells, the outer layer being called the *ectoderm*, the inner layer, the *endoderm.* Between the ectoderm and endoderm is a non-cellular substance called the *mesoglea.* Compare the size and shape of the cells in the two layers. What are the large clear spaces in the endoderm cells?

Make a drawing of a cross section through the stalk, showing the cellular details.

Exercise 49

OTHER COELENTERATES

Observe preserved specimens of corals, sea anemones, and jellyfishes. Note the general body plan. What kind of symmetry is shown? Identify the mouth and the tentacles. Like the hydra, all coelenterates have a single cavity, the coelenteron. Which forms studied are solitary and which are colonial? In which is an external skeleton present?

Make drawings of the forms studied.

Exercise 50

PLANARIA

Examine living specimens of *Planaria*, a common fresh-water flatworm. Locomotion is accomplished by cilia, which cover the body, and by muscular contraction. What kind of symmetry is shown? Observe (*m*) the distinct head with its two *eyespots*. Mount an animal on the underside of a cover glass that projects slightly beyond the edge of a slide. Study the ventral surface of the body (*lp*). Find the *mouth* near the middle of the body. It leads to a sac-like *pharynx*, which is everted as a proboscis during feeding. Just behind the mouth is the *genital pore*.

Study a stained specimen mounted on a prepared slide (*lp*). The pharynx is connected with an *intestine* that has three main branches. What is their position? Identify the bilobed *ganglion* situated between and below the eyespots and observe the two longitudinal *nerve cords* extending backward from it.

Draw an outline of the body (×10) and put in the digestive and nervous systems.

Exercise 51

EXTERNAL FEATURES OF EARTHWORM

Obtain a living or preserved earthworm and study its external features. Note that the body is composed of a series of ring-like segments called *metameres*. Compare the shape of the *anterior* (forward) and *posterior* (rear) ends of the animal and of the *dorsal* (upper) and *ventral* (lower) surfaces. Draw the body through the fingers and note the feel. Examine the ventral surface (*m*) and determine the number of rows and location of the bristle-like *setae*. Study (*m*) the anterior metameres and identify the *mouth* and *prostomium* (upper lip). Which metamere is incomplete? (The upper lip is not a metamere.) Locate the *anus* (*m*) at the posterior end of the body. The thick band-like swelling on the body wall is the *clitellum*. How many metameres does it cover? How many metameres do you count anterior to the clitellum?

Draw a ventral view of the anterior portion of the earthworm (×2), including two metameres posterior to the clitellum. Number every fifth metamere. Show all the external features observed.

Exercise 52

INTERNAL STRUCTURE OF EARTHWORM

Place a dead earthworm in a dissecting pan and cover with water. Fasten to the wax dorsal side up with a pin at each end of the body. Insert the point of the scissors in the body wall just behind the clitellum and slightly to one side of the dorsal blood vessel, which occupies a median position along the upper side of the body. Make a longitudinal incision through the body wall to the anterior end, working with great care so as not to injure the internal organs. While dissecting, observe that the body cavity, or *coelom*, is divided into compartments by transverse partitions called *septa*, each one corresponding to a metamere. After completing the longitudinal incision, cut through the septa with the point of a sharp scalpel, holding the edges of the body wall with the forceps. Pin the edges of the body wall to the wax as you proceed forward, slanting the pins outward.

First study the digestive system. The mouth opens into the large muscular *pharynx*, which can readily be identified. Behind the pharynx is the short tubular *esophagus*, somewhat hidden by the white male reproductive organs. The esophagus leads to the thin-walled *crop* and muscular *gizzard*, these two organs being in contact with each other. The long straight *intestine* extends posteriorly to the anus. Now study the circulatory system. You already have seen the *dorsal blood vessel*, which extends the entire length of the body. A similar vessel, the *ventral blood vessel*, occurs just beneath the digestive tract. Identify and count the number of pairs of *aortic arches* that pass around the esophagus, connecting the dorsal and ventral blood vessels. The smaller blood vessels will not be studied.

Make a drawing of the dorsal aspect of your dissected specimen (×2), showing what you have seen. The blood vessels may be drawn with red ink.

Exercise 53

BODY PLAN OF EARTHWORM

Obtain a prepared slide showing a cross section through the posterior portion of the body of an earthworm, and study (*lp*). Identify the body wall, the body cavity (*coelom*) and the digestive

tube (*enteron*). The dorsal wall of the intestine is infolded to form the *typhlosole*. Observe above the intestine the *dorsal blood vessel* and below it the *ventral blood vessel*. Just beneath the latter is the *ventral nerve cord*. Portions of the tubular excretory organs, the *nephridia*, may be seen in the coelom, and some of the setae, projecting through the body wall, may be present. Both the body wall and the wall of the intestine are composed of several different kinds of tissues, but muscle fibers predominate.

Make a semidiagrammatic drawing of the cross section studied, having the dorsal side of the animal toward the top of the page.

Exercise 54

EXTERNAL FEATURES OF STARFISH

Observe a preserved specimen of *Asterias* or of some other typical starfish. What kind of symmetry does it exhibit? Note the central *disk* and the five *rays*. On which side of the body are the spines longer? Find the *mouth* in the center of the oral surface. Extending outward from it into each ray is an *ambulacral groove* containing the *tube feet*, the organs of locomotion. How many rows of tube feet are present in each ray? Examine (*m*) the round, hard body on the aboral surface. This is the *sieve plate*. Observe (*m*) the *eye* at the tip of each ray.

Make a drawing of the oral and of the aboral surface of the starfish ($\times 1$), indicating the spines and other structural features on the disk and on one ray only.

Exercise 55

INTERNAL STRUCTURE OF STARFISH

Place a preserved specimen of a starfish in a dissecting pan, aboral side up, and cover with water. With scissors, cut through the body wall and around the edge of the ray directly opposite the sieve plate. Lift the flap slightly and with a scalpel carefully cut away the connections between the body wall and the internal organs. Observe the pair of *pyloric caeca* (digestive glands) which occupy the greater part of the ray cavity.

Now remove the entire aboral surface of the disk except the portion around the sieve plate and between the sieve plate and the center of the disk. Observe the *stomach*, the sac-like organ

that occupies the greater portion of the disk cavity. Note the short pouches extending from it into each ray. How are the pyloric caeca joined to the stomach? A small *intestine* leads from the stomach to the aboral surface but is difficult to see.

Draw an outline of the body and put in the digestive system.

Remove the digestive organs and study the water-vascular system. Find the *radial canal*, a small tube lying in the ambulacral groove. By cutting a ray transversely, note carefully its connection with the tube feet. At the upper end of each tube foot, observe (*m*) the small bulbous *ampulla*. Trace one of the radial canals to the disk where it joins the circular *ring canal*. Find the stone canal, a slender tube that connects the ring canal with the sieve plate.

Make a semidiagrammatic drawing of the water-vascular system of the starfish.

Exercise 56

OTHER ECHINODERMS

Examine specimens of brittle stars, sea urchins, sand dollars, and sea cucumbers, and compare with the starfish. What kind of symmetry do these animals have? Note the spiny skin (smooth in sea cucumbers), a feature that gives the echinoderms their name. How many rays are present in each case? By what are these organs represented in the sand dollars and sea urchins? Where is the mouth? Where are the tube feet?

Make drawings of the forms studied.

THE HIGHER INVERTEBRATES

Exercise 57

FRESH-WATER MUSSEL

Examine a specimen of a fresh-water mussel and study its external features. The soft body is enclosed by a calcareous shell consisting of two *valves* united dorsally by a *hinge ligament.* Observe on each valve a small rounded projection, the *umbo,* surrounded by lines of growth. Slightly force the valves apart and observe the soft *mantle,* a muscular sac that encloses the internal organs.

Remove the animal's left valve by inserting a scalpel and severing the two large *adductor muscles* that hold the valves together. Study the organs lying in the mantle cavity. Note the thick muscular *foot* at the anterior end. At the posterior end identify the *dorsal* and the *ventral siphons.* Observe the sheet-like *gills* over which water passes as it enters the mantle cavity through the ventral siphon. How many gills occur on each side?

With a pair of scissors remove the left mantle lobe and study the digestive system. Find the *stomach* and the large greenish *digestive gland* that surrounds it. A short *esophagus* extends from the stomach to the mouth. The latter is very small and is surrounded by four *palps.* Trace the long coiled *intestine* from the stomach to the anus, noting its passage through the heart. The *heart,* enclosed by a *pericardium,* consists of a dorsal *ventricle* and two dorsal *auricles.* Identify these. Trace the anterior artery and the posterior artery as far as you can. Observe the pair of brown spongy *kidneys* below the heart.

Draw an external view of the mussel as seen from the side (×1). Make a drawing of the internal organs in place in the right valve (×1).

Exercise 58

OTHER MOLLUSKS

Study specimens of clams, oysters, and scallops, noting their resemblances to the mussel. In which forms are a foot and

siphons absent? In the scallop note the numerous slender tentacles and row of eyes borne by the mantle folds.

The snails have univalve shells while the related slugs have none. In what other ways do these forms differ from the bivalve mollusks? What is the function of the foot? Compare the symmetry of a snail with that of a mussel. How do you account for the difference?

Examine an octopus and a squid. Notice the head and eyes. The foot is divided into tentacles, which surround the mouth. How many are present? What other peculiarities do these animals exhibit?

Based on your observations and the above questions, write an account of your study.

Exercise 59

EXTERNAL FEATURES OF CRAYFISH

The body of the crayfish is composed of a *head*, *thorax*, and *abdomen*, but the head and thorax are fused to form a *cephalothorax*. Observe the hard plate, called the *carapace*, which covers the top and sides of the cephalothorax. Note the *cervical groove* on its dorsal surface, marking the boundary line between the head and thorax, and the *rostrum*, a beak that projects from the anterior end of the carapace. On either side of the rostrum is a large *compound eye*, located at the end of a movable stalk.

Study the ventral surface of the animal, noting the metameric arrangement of the 19 pairs of appendages. Beginning anteriorly, identify the pair of short *antennules*, the pair of long *antennae*, six pairs of *mouth parts*, one pair of *chelipeds* (pincers), four pairs of *walking legs*, five pairs of *swimmerets*, and one pair of *uropods*. The last abdominal metamere, the *telson*, lacks appendages and forms, with the uropods, the fan-shaped tail. Observe and describe the difference between the male and female with respect to the first two pairs of swimmerets.

Draw a dorsal view of the crayfish (×1) showing all the features you have observed.

Exercise 60

APPENDAGES OF CRAYFISH

All the appendages of the crayfish, despite their superficial differences, show the same fundamental plan of organization.

Study the structure of the right swimmeret borne by the fourth abdominal metamere. Note that it is composed of three main divisions, the basal portion being the *protopodite*, the outer branch the *exopodite*, and the inner branch the *endopodite*. Identify these divisions and then remove the swimmeret.

Now remove the right member of each of the following pairs of appendages and identify their divisions: uropods, fourth walking legs, second maxillipeds (the fifth pair of mouth parts), second maxillae (the third pair of mouth parts), the antennae, and the antennules. The chelipeds and walking legs have no exopodite; of their seven joints, the lower two constitute the protopodite. The protopodite of the second maxilla is broad and leaf-like, while the endopodite is small and the exopodite broad. The endopodite of the antenna is jointed and very long, the exopodite plate-like and short.

Make a careful drawing of each of the appendages removed (×1), labeling each division.

Exercise 61

INTERNAL STRUCTURE OF CRAYFISH

Place your specimen in a dissecting pan and cover it with water. With the scissors cut away the lateral portion of the carapace and expose the *gills*. Observe their number, structure, and place of attachment. Now remove the dorsal portion of the carapace, being careful not to injure the internal organs. Remove the membrane from the head region and observe the *stomach;* it is situated just back of the rostrum. Notice that it is composed of an anterior portion (*cardiac chamber*) and a posterior portion (*pyloric chamber*). It is connected with the mouth by a short *esophagus*.

Just back of the stomach, enclosed by a transparent membrane called the *pericardium*, is the *heart*. Five delicate arteries arise from its anterior end. Find them and trace them forward as far as possible. Carefully press aside the heart and observe the artery that arises near its posterior end and goes downward, giving rise to an anterior and a posterior branch. Also note the artery that arises from the posterior end of the heart and goes backward along the dorsal abdominal wall. Remove the heart and place it in some water. Observe (*m*) its three pairs of lateral openings.

Identify the pair of large pale-greenish-yellow *digestive glands* on both sides of the stomach. Sexual organs will be found here too. In the female find the large, three-lobed yellow or brown *ovary*. In the male find the slender, white, three-lobed *testis* and observe the slender, convoluted cords (*sperm ducts*) attached to it. Trace them to their outlet at the base of the last pair of walking legs. In the female trace the pair of *oviducts* from the ovary to their outlet at the base of the second pair of walking legs. Trace the *intestine* from the stomach to the anus, dissecting away the muscles of the abdomen. Find the pair of *green glands*, the organs of excretion, which lie in the head region below and in front of the stomach. Trace their connection with external openings on the basal joint of the antennae.

Draw a side view of the digestive system and an outline of the body around it (×1). Also indicate the form and position of those parts of the circulatory, reproductive, and excretory systems seen.

Remove all the muscles and other organs from the body and find the *ventral nerve cord* lying along the mid-ventral line of the abdomen. Trace it forward into the cephalothorax, using great care in dissection. How many ganglia do you find in the abdomen and how many in the cephalothorax? Find the *subesophageal ganglion* immediately back of the esophagus and the *supra-esophageal ganglion* just back of the eyes. Note how these ganglia are connected.

Add the nervous system to the drawing previously made.

Exercise 62

MYRIAPODS

Examine specimens of centipedes and millipedes, noting the worm-like form of the body with its distinct head, but no differentiation into thorax and abdomen. How do the centipedes and millipedes compare as to shape of the body and number of legs per metamere? Note (*m*) the single pair of *antennae*, the *mouth parts*, and the *eyes*. In most myriapods two *simple eyes* are present, but some millipedes have a group of simple eyes on each side of the head, each group constituting an *aggregate eye*. In the centipede note the large hook-like *maxillipeds* just back of the head and the *anal feelers* at the posterior end of the body.

Draw a dorsal view of a centipede (×2) and a side view of a millipede (×1 or larger).

Exercise 63

EXTERNAL FEATURES OF GRASSHOPPER

The body of a grasshopper, like that of all insects, is made up of a *head*, *thorax*, and *abdomen*. Identify these divisions in the specimen provided for this exercise. In which is metamerism most apparent? To which division are the legs and wings attached?

Study (*m*) the pair of large *compound eyes*, the pair of long jointed *antennae* (feelers), and the three simple eyes or *ocelli* arranged in the form of a triangle on the front of the head. On the ventral side of the head are the *mouth parts*. With a razor blade make a thin slice through one of the compound eyes, prepare a temporary mount, and note (*lp*) that it is composed of numerous small *facets*.

The thorax of the grasshopper is composed of three metameres, termed the *prothorax*, *mesothorax*, and *metathorax*. Each bears a pair of legs. Spread out the two pairs of wings and determine to which division of the thorax they are attached. In what ways do the fore and hind wings differ? Study one of the metathoracic legs. Of how many parts is it composed? Find (*m*) the two pairs of thoracic *spiracles* (breathing pores). On what metameres are they borne?

The abdomen is composed of 10 metameres. The first one is incomplete. Identify (*m*) the pair of *auditory organs* that it bears. How many of the abdominal metameres bear spiracles? In the female the tip of the abdomen is modified to form an egg-laying organ termed the *ovipositor*, that of the male to form an organ for transferring sperms. Observe both sexes and compare.

Draw a side view of the grasshopper with wings raised (×2). Also draw a portion of a compound eye.

Now study the mouth parts. With the forceps lift the *labrum* (upper lip) and observe the pair of hard black *mandibles* (biting jaws) beneath it. Remove both sets of mouth parts and examine (*m*). Then remove the remaining mouth parts, consisting of the *first maxillae*, *second maxillae* and *hypopharynx* (tongue). The second maxillae are united laterally to form the *labium* (lower lip).

Make an accurate drawing of the mouth parts (×4).

Exercise 64

INTERNAL STRUCTURE OF GRASSHOPPER

Select a large female grasshopper for dissection. Cut off the wings and legs close to the body and pin the specimen to the wax in a dissecting pan with the dorsal side up. Cover with water. Make a cut with the scissors along each side of the dorsal body wall of the abdomen and very carefully remove it. Do not injure the internal organs. Dissect forward and remove the body wall of the thorax and head. Observe (*m*) the *tracheae*, small branched tubes that extend throughout the entire body, carrying air to the tissues. In the posterior part of the abdomen find the pair of large *ovaries*. They may contain many eggs. Trace (*m*) the connection between the ovaries and the ovipositor.

Remove the left lateral wall and study the digestive system. Identify the short curved *esophagus* leading from the mouth to the *crop*. The crop is a large organ extending from the mesothorax to the anterior part of the abdomen. Back of the crop is a small *gizzard* which opens directly into a large thin-walled *stomach*. The stomach extends from the first to the seventh metamere of the abdomen. Trace the *intestine* from the stomach to the anus. At the anterior end of the stomach observe the large *gastric caeca* (digestive glands). What are their number and arrangement? At the posterior end of the stomach find the ring of very delicate *Malpighian tubes*, the excretory organs.

Make a drawing of a side view of the grasshopper (×3), showing all the internal organs seen.

Exercise 65

OTHER INSECTS

Examine insects of the following kinds: a dragonfly, a butterfly, a bug, a fly, a beetle, and a wasp. Make a list of all the features that these forms have in common. Then note their differences, comparing them as to the number and character of the wings, the nature of the mouth parts and the functions to which they are adapted, and any special features that you are able to observe.

Make a drawing of each insect studied (×1 or ×2).

Exercise 66

ARACHNIDS

Examine a large spider, noting the division of the body into *cephalothorax* and *abdomen.* Is metamerism evident? Do you find antennae, compound eyes, or wings? Note (*m*) the conspicuous mandibles and the maxillae, the latter bearing jointed palps for grasping food. How many simple eyes are present? Study their distribution (*m*). How many pairs of legs are present? To what part of the body are they attached? Observe (*m*) the pair of slits in the ventral side of the abdomen; they communicate with the respiratory system. At the tip of the abdomen note (*m*) the three pairs of *spinnerets* and the *anus.*

Draw a ventral view of the spider (×2). Also make a drawing of a front aspect of the cephalothorax (×4).

Observe specimens of scorpions, daddy longlegs, and ticks, noting in what ways they resemble spiders. In which forms is metamerism evident? The abdomen in the scorpions is differentiated into two portions and bears a terminal sting.

THE VERTEBRATES

Exercise 67

EXTERNAL FEATURES OF FROG

Study the living frog and write an account of your observations. What are the body divisions? In a typical vertebrate these are the head, trunk, and tail. Is the skin smooth or scaly? What is the relative length of the fore and hind limbs? Of how many divisions does each consist? What is the position of the limbs when the animal is at rest? Describe locomotion both on land and in the water. What is the function of the fore limbs? How are the hind limbs adapted for jumping and how for swimming? How many toes are there on each foot? Which ones are webbed? Are claws present on any of them? Note the number and position of the eyes, nostrils, and eardrums. Are the eyes movable? How many eyelids are present? Describe the movements of the nostrils, throat, and sides of the body. With what function are these movements concerned?

Make a side view of the frog (×1) showing all the external features observed.

Exercise 68

INTERNAL STRUCTURE OF FROG

Obtain a freshly killed frog and place it in a dissecting pan. Open the mouth wide and observe the fleshy *tongue*. Pull it forward with the forceps, noting the manner of its attachment and its extensibility. On which jaw do you find *teeth*? Observe the two small groups of teeth on the roof of the mouth. Locate the nasal openings on either side of them. Observe the slit-like *glottis* behind the tongue and just below the *esophagus*. It communicates with passages leading to the lungs.

Make a drawing of the mouth cavity (×2).

Place the animal on its back in the dissecting pan, cover with water, and pin to the wax through the tip of the jaws and through each of the four feet. Lift the skin with the forceps and make an incision with the scissors along the entire mid-ventral line of the

body. Cut outward at each end of this incision and pin back the folds of skin. Now cut through the ventral body wall as far as the middle of the lower jaw, cutting through the shoulder girdle. Be careful not to injure the internal organs. Cut transversely on either side and pin back the flaps. In the breeding season the ovaries will occupy a large part of the body cavity of the female, and it will be necessary to remove them before studying the other organs.

First study the digestive system. Observe the large, reddish, three-lobed *liver* and find between its lobes the small green *gall bladder*. Lift the liver and note carefully the size, shape, and location of the *stomach*. Insert a probe through the mouth and observe the short *esophagus*. Trace the course of the *small intestine*, noting that it is connected at its posterior end with the *large intestine*. Find the *pancreas*, a white irregular body lying between the stomach and the anterior end of the small intestine. Locate the small, globular, red *spleen* near the anterior end of the large intestine. Trace the large intestine to the *cloaca*, a wide cavity between the legs. The digestive system ends at the *anus*.

Turn the small intestine to one side and note the pair of elongated red *kidneys* lying close to the dorsal body wall. Trace their ducts, the *ureters*, to the cloaca and note its relation to the sac-like *bladder*. At the anterior end of the kidneys is a pair of oval yellow *testes* in the male and two irregular sac-like *ovaries* in the female. The latter become greatly distended during the breeding season. Note the finger-like *fat bodies* attached to the anterior end of the sex organs. Trace the delicate tubular *sperm ducts* from the testes to the kidneys. The pair of long coiled *oviducts* are not attached to the ovaries, but open freely into the coelom on either side of the esophagus.

Insert a slender tube into the glottis and inflate the *lungs*, noting their general appearance and position in the body cavity. The lungs are connected with the glottis by means of a pair of *bronchial tubes*. Study the *heart* lying within a delicate transparent sac (the *pericardium*). Identify the single *ventricle* and the *right* and *left auricles*. The ventricle is posterior to the auricles. Note that the arterial system arises from the anterior end of the ventricle on its right side.

Make a large drawing of the dissection as far as you have gone, showing as many organs as possible. Also make a ventral view of the heart (×3).

Exercise 69

BRAIN OF FROG

Remove the flesh from the back of the neck and head. Bend the head down and find a small space about 3 mm. long at the junction of the skull and vertebral column. Remove the membrane covering it and observe the *brain* lying below. With the scissors make a slit from this space along each side of the head to the eyes. Lift off the roof of the skull with the forceps and very carefully remove the dark-colored material above the brain. At the anterior end observe the pair of elongated *cerebral hemispheres* prolonged anteriorly into the *olfactory lobes.* Identify the pair of oval *optic lobes* and behind them the small narrow *cerebellum.* At the posterior end of the brain is the triangular *medulla oblongata,* which merges into the *spinal cord.* Note that nerves are given off both from the brain and from the spinal cord.

Make a dorsal view of the brain (×4).

Exercise 70

SKELETON OF FROG

Study a prepared dried skeleton of a frog and identify the *skull, vertebral column, limb girdles,* and *limbs.* Count the *vertebrae;* note the pair of slender transverse processes arising from each. The fore limbs are attached to the *pectoral girdle,* the hind limbs to the *pelvic girdle.* Distinguish between the *upper arm, forearm, wrist,* and *hand.* How many *fingers* are there? Identify the *thigh, shank, ankle,* and *foot.* How many *toes* are present?

Make a diagram of the frog's skeleton (×1), showing its main parts.

Exercise 71

LATER DEVELOPMENT OF FROG

Examine a series of frog tadpoles (of the same species) arranged according to age. In the youngest stage note the external gills and the absence of eyes and mouth. In the later stages observe the gradual development of the limbs and the disappearance of the tail.

Draw at least four stages in the later development of the frog.

Exercise 72

OTHER VERTEBRATES

Fishes.—Study living or preserved specimens of fishes. Note the character of the body covering. How many paired fins are present? How many unpaired fins do you find? Which fins are comparable to the fore and hind limbs of the higher vertebrates? Fishes breathe by means of gills which, in all except the lowest forms, are covered by a flap. Note the movements concerned with breathing, if the specimen is alive.

Amphibians.—Observe specimens of frogs and toads and of salamanders and newts. Is the skin smooth or covered with scales? How many limbs are present, and how many toes on each? Which forms have a tail in the adult stage of development? When young all amphibians breathe with gills; in some cases these are retained throughout life, but in most of them they are replaced by lungs.

Reptiles.—Examine a lizard, a snake, a turtle, and an alligator. What is the character of the skin? How many limbs are present in each case? Do the toes bear claws? How many toes occur on each foot? Do reptiles breathe with gills or with lungs when young? What special features do you observe in the turtle?

Birds.—Nearly any available kind of bird will serve to illustrate the general characteristics of this class of vertebrates. While the skin on most parts of the body bears feathers, where do you find scales? To what organs in other vertebrates are the wings comparable? Are the toes clawed? What are their number and arrangement? What are the peculiarities of the jaws in birds?

Mammals.—Almost any typical mammal will be satisfactory to examine as a representative of the highest group of vertebrates. What is the characteristic body covering in the mammals? Are the fore limbs as highly modified as in the birds? In the form studied do the toes end in claws, nails, or hoofs? How many toes are present on each foot? How do the jaws differ from those of birds? Are mammals cold blooded or warm blooded? Compare with the other classes of vertebrates in this respect. How are the young nourished?

METABOLISM AND IRRITABILITY IN ANIMALS

Exercise 73

TESTS FOR FOOD SUBSTANCES

After making each of the following tests, describe the method of procedure and state the results obtained.

Starch.—Place a piece of starch the size of a pea in a test tube, fill two-thirds full of water, and shake. Add a few drops of iodine solution (iodine crystals dissolved in a 2 per cent aqueous solution of potassium iodide), and note the color reaction.

Sugar.—Fill a clean test tube about one-quarter full of Fehling's solution,[1] bring to a boil, and add a few drops of a solution of grape sugar[2] in water. Reboil if no color change is immediately apparent.

Protein.—Place a very small amount of raw white of egg in a test tube, fill half full of water, and shake. Add about 2 cc. of a strong solution of potassium hydroxide, and boil for a few seconds. Then add a few drops of a strong solution of copper sulphate, shake well, and observe the color reaction.

Fat.—Place a small quantity of corn meal or of other food containing fat in a test tube, fill half full of ether, and shake. Carefully pour off the liquid in a flat dish and place outside the window until the ether has evaporated. What is left?

Exercise 74

DIGESTION

Place a piece of starch the size of a pea in a test tube, fill two-thirds full of water containing the enzyme diastase[3] (1

[1] Take equal parts of a 1.5 per cent solution of cupric sulphate, an 8 per cent solution of sodium potassium tartrate, and a 6 per cent solution of sodium hydroxide. These solutions can be prepared in advance but should not be mixed together until needed for use.

[2] If cane sugar is used, add 2 or 3 drops of dilute sulphuric acid (1 part sulphuric acid to 5 parts water) to 5 cc. of the sugar solution and boil. Then proceed as with the grape sugar.

[3] Or use tap water and add about 5 cc of saliva, which contains ptyalin, another starch-digesting enzyme.

gram of dry diastase of malt to 1,000 cc. of water), and shake. In another test tube place several small pieces of hard-boiled white of egg, and fill two-thirds full of water containing hydrochloric acid and the enzyme pepsin (1 gram of dry pepsin to 1,000 cc. of a 0.2 per cent solution of hydrochloric acid).

Put both tubes in a water bath or in an incubator and keep at a temperature of approximately 40°C. After several hours observe the appearance of the material in the tubes. What happens to the starch and protein? Test the liquid in the first tube for starch and sugar (see Exercise 73).

Describe the experiments and write a statement as to just what the process of digestion involves.

Exercise 75

RESPIRATION

By means of a piece of glass tubing, blow into a test tube containing limewater. Note the formation of a white precipitate. The carbon dioxide exhaled from the lungs has combined with the calcium hydroxide dissolved in the water to form calcium carbonate, which is insoluble. Now attach a large rubber bulb to the glass tubing and blow air into fresh limewater. The atmosphere contains approximately 0.03 per cent of carbon dioxide. Is this amount sufficient to affect the limewater?

Briefly describe the experiment.

Exercise 76

CIRCULATION

A frog will be anesthetized and prepared so that you may study under the microscope the circulation of blood in the web of the hind foot. Examine (*lp*) and note the pigment spots and blood vessels. How can you distinguish between the *arteries* and *veins?* The smallest vessels, called *capillaries*, connect arteries and veins or may anastomose and connect one vein with another. Note carefully the movement of the blood corpuscles and describe. The corpuscles are carried along in the colorless *plasma.*

Draw a portion of the web showing arteries, veins, and non-anastomosing capillaries.

Exercise 77

REFLEX ACTION

The frog used in this experiment has had its brain destroyed by means of a stout wire passed through the base of the skull. It is entirely unconscious. Suspend the animal from a support so that the head is uppermost. During the experiment keep the skin moist. With the forceps dip a piece of filter paper about 2 mm. square in dilute acetic acid and place it on the frog's abdomen. Note the nature of the response, determining the number of seconds which elapse between the application of the stimulus and the reaction. Since the brain has been destroyed, how is the response controlled? Trace the path of the stimulus through the parts of the body involved. Why is this behavior called reflex action?

Exercise 78

IRRITABILITY

For this exercise a number of specimens will be provided of *Planaria*, a common fresh-water flatworm. Touch one of the animals with the point of the forceps and observe its reaction. What is the nature of the stimulus? Observe animals placed in a dish that is partly darkened by means of black paper. What position do they assume with reference to the light? In the dish containing vigorous green algae, where are most of the flatworms? What is the nature of the stimulus involved in this case? With a dropping pipette place a very little meat juice near the animals, or place a small piece of liver in the culture dish. Note the behavior of the animals and explain the nature of the stimulus.

Write an account of your observations and explain behavior seen.

REPRODUCTION AND DEVELOPMENT IN ANIMALS

Exercise 79

SEXUAL REPRODUCTION

Examine (*m*) eggs of the frog, or of any other amphibian with similar breeding habits, and of any species of oviparous fish. Observe the size of the eggs, their approximate number, their covering, and the way in which they are held together in a mass. These eggs, produced in the ovaries of the female, are fertilized while or after being laid. The embryos develop in the water.

Examine eggs of a reptile and of a bird. The yellow portion, or *yolk*, is the true egg, the white part being accessory food material. Why are these eggs provided with a hard shell, while those of fishes and amphibians are not? Where are they fertilized?

Obtain a prepared slide showing sections through an ovary with unfertilized eggs in various stages of development. Study the eggs (*hp*), noting the conspicuous nucleus and relatively large amount of cytoplasm. If the ovary of a mammal is studied, note that each egg is surrounded by a number of small *follicle cells*.

Examine newly laid eggs of pond snails (*m*) and observe that on the surface of each egg may be seen two or three small round *polar bodies*. By two successive cell divisions, the immature germ cells (*oögonia*) give rise to four cells, one of them being comparatively large and three small.

Draw and describe all types of eggs studied.

With the dissecting needles break up on a slide in a normal salt solution (a 0.6 per cent solution of sodium chloride) a small piece of the testis of a recently killed frog, mount, and examine (*hp*). Observe and draw the living sperms, noting the rod-like, slightly curved head and the long slender tail. Each one is a single cell.

Exercise 80

EARLY EMBRYONIC DEVELOPMENT

Study a prepared slide (*hp*) showing starfish or sea urchin eggs in various stages of *cleavage*. Draw two-, four-, and eight-celled stages and at least one later stage. As the process of cleavage progresses, how do the cells compare in size?

Examine another slide showing stages in *gastrulation*. Find and draw a *blastula*, which consists of a single layer of cells surrounding a cavity (*blastocoele*). The *gastrula* develops by a bulging inward of the cells on one side of the blastula. Observe this feature and draw an early and a late stage in gastrulation. The new cavity (*gastrocoele* or *archenteron*) is surrounded by two layers of cells, the outer one being the *ectoderm* and the inner the *endoderm*. In the form studied the blastocoele is not obliterated by the process of gastrulation, as it is in many other cases. What animal that has been studied goes no farther in its development than the gastrula stage?

Older stages will show *mesoderm* cells arising between the ectoderm and endoderm. Observe this feature and draw.

Exercise 81

CHIEF ANIMAL TISSUES

Arising from undifferentiated cells of the embryo, various types of tissues appear during the course of development. The chief ones will be briefly studied here.

Covering the external and internal surfaces of organs is a relatively simple type of tissue called *epithelium*. Examine (*hp*) and draw whatever kinds are available, noting the shape and arrangement of the individual cells.

Study a prepared slide showing *smooth muscle cells*, and note (*hp*) their elongated character. What is the shape of the nucleus and the ends of the cell? Examine (*lp*) prepared longitudinal sections of *striated muscle*, and note the bundles of muscle fibers.

Note (*hp*) that each fiber is a very long multinucleate cell with numerous cross striations.

Draw several smooth muscle cells and a portion of a striated muscle fiber.

Examine preparations showing isolated *nerve cells*. Distinguish carefully (*hp*) between the *cell body* and the *nerve fibers*. Note the shape of the former and any differences between the latter.

Draw several nerve cells showing the features observed.

Study (*hp*) a prepared slide of sectioned *cartilage*, noting the cartilage cells and the intercellular matrix. How are the cells arranged? Study (*hp*) prepared cross sections of dry *bone*. The small black areas are spaces formerly occupied by living bone cells. Note the wavy canals radiating out from them; through these there extended slender protoplasmic extensions of the bone cells. Observe carefully the structure of the matrix. In both cartilage and bone what can you infer as to the origin of the matrix?

Make drawings of cartilage tissue and of bone.

Exercise 82

INSECT METAMORPHOSIS

Examine a *larva*, *pupa*, and *adult* of the same species of butterfly or moth. The egg hatches into a worm-like caterpillar that leads an independent life. This represents the larval stage. Note its metameric body composed of a distinct head, thorax, and abdomen. Observe the difference in structure between the three pairs of thoracic legs and the abdominal legs. How many pairs of the latter are present? What organs are present on the head?

Eventually the larva goes into a resting condition, becoming a pupa. In the form studied does the larva spin a silken *cocoon* within which it pupates? What organs can you recognize in the pupa? Profound changes go on during the pupa stage, and finally the insect emerges as a full-fledged adult. Identify the three body divisions of the adult, the large compound eyes, the antennae, the fore and hind wings, and the three pairs of legs. Upon which part of the body are the wings and legs borne?

Make a drawing of each of the three stages in the development of a butterfly or moth.

PHYSICAL BASIS OF HEREDITY

Exercise 83

MITOTIC CELL DIVISION

Prepared slides of root tips (or of other favorable material) showing various stages in vegetative mitosis will be found under the demonstration microscopes. Study each one very carefully before drawing. Because of the high magnification, it will be necessary to focus slightly with the fine adjustment as you study the cells. This is important, for without focusing you will be unable to see all the details.

In the early stages of mitosis, constituting the *prophase*, note the presence of numerous deeply stained rod-shaped bodies formed by the consolidation of the chromatin of the resting nucleus. These are *chromosomes*. Is the nuclear membrane still present and can the nucleolus be identified?

In a later stage, the *metaphase*, a bipolar spindle formed of very delicate fibers can be seen, the chromosomes being arranged at the equator. Because you are looking at the spindle from the side, the chromosomes seem crowded together, but several of the individual ones may usually be seen by careful focusing. Note that each chromosome is longitudinally split in two.

After the split chromosomes have become arranged at the equator of the spindle, the halves of each one move toward opposite poles. Observe the stage showing the two groups of separating chromosomes, the *anaphase*, and note that most of them are U-shaped. After the two groups of chromosomes have reached the poles, a cell wall forms between them on the spindle fibers, thus cutting the old cell in half. Observe such a *telophase*, noting the condition of the chromosomes. In the case of animal cells the cell is separated by a constriction of the cytoplasm.

Make a series of very accurate drawings showing all the stages in mitosis that you have studied. Also draw a single resting cell.

Exercise 84

SPERMATOGENESIS

Examine (*lp*) a prepared slide showing longitudinal sections of a testis of a mature salamander or a grasshopper. All stages in the development of sperms should occur on the slide. The immature germ cells (*spermatogonia*) undergo mitotic cell division and increase in number. Finally they stop multiplying and increase in size. Note (*hp*) these larger cells (*primary spermatocytes*) and study. Make a drawing of one of them. In some the nucleus is in the resting condition; in others it is undergoing the first reduction division. At this time the chromosomes do not split longitudinally, as in an ordinary mitosis, but come together in pairs, a behavior known as *synapsis*. Find a favorable cell, count the number of chromosome pairs, and draw. How many whole chromosomes will go to each of the resulting cells (*secondary spermatocytes*)? How does this number compare with the number of chromosomes present in all the somatic cells and in the unripe germ cells? (The instructor will tell you the diploid chromosome number for the species you are studying.)

A second division results in the formation of four small cells (*spermatids*) that metamorphose into sperms. This division represents an ordinary mitosis. Draw a group of mature sperms, noting the small head and long slender tail.

In addition to making the detailed drawings called for above, which should be very accurate, make a diagram to represent the process of spermatogenesis in the form studied.

MENDELIAN LAWS OF HEREDITY

Exercise 85

PRINCIPLE OF SEGREGATION

When starchy corn is crossed with sweet corn, all the seeds of the hybrid offspring are starchy. Starchiness is therefore the *dominant* character, sweetness the *recessive*. When the hybrids are interbred, however, both kinds of seeds make their appearance. Examine ears of corn produced by such hybrids. Count the number of each kind of grains in four rows on the ear. What proportion of them are starchy (plump) and what proportion sweet (shriveled)? How does this compare with the theoretical ratio that would be expected on the basis of Mendel's principle of segregation? If you were to remove some seeds of each kind from the ear you are studying, plant them, and self-fertilize each of the resulting plants, what kind of offspring would you expect from the starchy corn? From the sweet corn? Would all the starchy corn behave alike under these conditions? Explain.

Write a complete explanation of this experiment in terms of Mendelian heredity.

Exercise 86

PRINCIPLE OF SEGREGATION (*Cont'd*)

For this experiment you will be provided with a bottle containing a pair of fruit flies (*Drosophila*) and a piece of fermenting banana.[1] One fly has long (normal) wings, the other vestigial wings. Determine which is the male and which the female[2] and

[1] To prepare cultures use clean, 8-ounce, wide-mouth bottles stoppered with cotton plugs. Into each put a small piece of ripe banana, and sterilize in an autoclave. Mix up some compressed yeast in water and put about 10 drops on the banana. Add a folded piece of filter paper to absorb surplus moisture. Cultures should be kept at a temperature of about 25° C. To obtain virgin females, remove all adults from a culture containing pupae, and isolate the females within 6 hours of the time of their appearance.

[2] The female is larger than the male and has a pointed abdomen. The abdomen of the male is blunt and rounded and black on the underside.

which parent has the long wings. Record these data on the label together with the date when the flies were mated. The female will lay a great many eggs, which will soon hatch into white larvae. On or about the tenth day of the experiment release the parents. The brown inactive pupae will begin to appear about this time, and soon thereafter the first adults will emerge. These flies are hybrids. Which of their parents do they resemble? Is the vestigial wing character *dominant* or *recessive?*

Remove the hybrid flies from the culture by taking out the cotton plug and quickly placing an empty bottle over the open neck. Point the empty bottle toward a window and the flies will move toward the light. Now pour a little ether on a new cotton plug and place it in the neck of the bottle containing the adult flies. Allow 20 to 30 seconds to elapse. After the flies have become etherized they can be placed on a piece of paper and easily handled with a small camel's-hair brush. Obtain a fresh culture bottle and place within it one or two pairs of the hybrid flies, allowing them to drop on the filter paper. After about 2 weeks the second-generation flies will begin to appear. Remove these flies as before, and count the number of long-winged and of vestigial-winged individuals. What is the actual number and the relative proportion of the two kinds of offspring that the first-generation hybrids have produced? What proportion of the long-winged flies are themselves hybrids? What kind of a cross would you make in order to determine whether a given fly is pure (*homozygous*) or hybrid (*heterozygous*) for the long-winged condition? Explain.

Fully describe the experiment and write a complete explanation of the results obtained.

ADAPTATION

Exercise 87

XEROPHYTES

Xerophytes are plants that live in dry places. They are structurally adapted to live under conditions of little available moisture. Observe, in the field if possible, various types of xerophytes, such as those with reduced leaves, with hairy leaves, with stiff or leathery leaves, and with succulent leaves or stems. Examine (*hp*) cross sections of a leaf of one or more of the plants studied, and note all the ways in which its internal structure differs from that of an ordinary leaf.

Write an account of your observations, stating in each case exactly how the plant is adapted to endure the conditions under which it lives. Explain fully the significance of all of the adaptive features seen.

Exercise 88

SEED DISPERSAL

Examine winged seeds[1] of catalpa, maple, ash, and elm and downy seeds of milkweed, dandelion, and thistle. In each case note the development of structures to facilitate dispersal by the wind. Examine the barbed fruits of beggar-ticks and the heads of cocklebur and burdock. Study (*m*) the individual barbs. How are these seeds adapted for dispersal?

Draw an example of each type of dispersal seen.

Exercise 89

PROTECTIVE RESEMBLANCE

Observe mounted specimens of insects showing protective resemblance, such as the katydid, walking stick, dead-leaf

[1] Except in the case of the catalpa and milkweed, the "seeds" are really small, dry, one-seeded fruits, but this distinction has no significance from the standpoint of dispersal.

butterfly, underwing moth, etc. Point out the ways in which these animals harmonize with their surroundings. Of what value is this to the animal itself?

Exercise 90

FEET AND BILLS OF BIRDS

Compare the feet and bills of the following birds (or equivalent types): (*a*) The duck, a swimming bird which gets its food from the mud by digging with its bill. (*b*) The avocet, a wading bird which uses its bill to probe among stones along shores. (*c*) The woodpecker, a woodland bird which drills into tree trunks for grubs and other insects. (*d*) The hummingbird, a nectar feeder which poises on the wing before flowers, thrusting its long bill into the corolla. (*e*) The hawk, a flesh eater which seizes living prey with its feet. (*f*) The sparrow, a seed-eating bird which finds its food by scratching.

Describe the ways in which each type of bird is adapted to its peculiar life habits. Make a sketch of the head and foot of each bird studied.

Exercise 91

TEETH OF MAMMALS

Examine the skull of a man or a monkey and identify the four kinds of teeth: *Incisors*, *canines*, *premolars*, and *molars*. In man there are, in each jaw, four incisors, two canines, four premolars, and six molars. How many teeth are there in the complete set? Now examine the teeth of other mammals, such as those of a rodent, a carnivore, an ungulate, and an insectivore. In each case tell the number of each kind of teeth present and how they are specialized for particular functions.

Exercise 92

MAMMALIAN FORE LIMBS

A typical mammalian fore limb consists of an upper arm composed of a single bone, the *humerus*, a forearm composed of two bones, the *radius* and *ulna* (the latter forming the elbow), a wrist or *carpus*, and five *digits*. Identify these parts of the skeleton of man or another primate, and draw. Now compare

the structure of the fore limbs of a series of different mammals, such as the mole, dog, goat, seal, and bat, stating all the ways in which they differ from the human fore limb. Which do you regard as the most highly specialized? Why? How do you explain the general resemblance between the structure of the fore limbs in the different forms studied, that is, the fact that they are all constructed according to the same pattern? With what are the differences between them correlated?

PARASITISM AND SYMBIOSIS

Exercise 93

PARASITIC PLANTS

Lilac Mildew.—Examine a lilac leaf infected with lilac mildew (*Microsphaera*) and observe (*m*) the white thread-like body of the fungus (*mycelium*) growing upon its surface. How does it compare with that of the bread mold previously studied? Short branches of the mycelium penetrate the epidermal cells of the leaf and absorb nourishment from them. The spherical black bodies (if present) are reproductive structures; they contain spores. Why is this plant a parasite?

Make a sketch of the infected leaf with the mildew growing upon it.

A Rust Fungus.—Examine a rust-infected leaf of sunflower, snapdragon, mallow, or some other plant, noting the brown pustules on the surface. These are filled with spores of a rust fungus, the mycelium of which lives inside the leaf. As in the lilac mildew, the rust absorbs its food directly from the cells of the leaf, thus causing the host to become diseased. The mildew is an external or superficial parasite, while the rusts are internal parasites. In which case would the application of a fungicide be effective? Why? Make a sketch of the infected leaf.

Mistletoe and Dodder.—These are parasites belonging to the seed-plant group. They attack a number of different kinds of plants. Describe the way in which the parasite is attached to the host. Does the parasite have leaves, and if so are they conspicuous or reduced? In either case explain their presence. Does the parasite have any connection of its own with the soil? How does it obtain its water?

Make drawings of the forms studied, showing a portion of the host as well as the parasite.

Exercise 94

PARASITIC ANIMALS

Tapeworms are parasites that live in the digestive tract of the higher animals. Tapeworms from the dog or cat (as well as

other intestinal parasites) are easily obtained. Examine a preserved specimen, noting the series of flat segments. At the smaller end try to find the "head" (*m*), which serves as a means of attachment to the intestinal wall of the host. It is often broken off in removing the worm. The "head" gives rise to the posterior segments by a process of budding, and thus the worm is not a single individual but a colony.

Make a drawing of at least a portion of the specimen studied.

Exercise 95

SYMBIOSIS

Lichens.—Examine several different types of lichens, noting their color, general form, texture, and growth habit. A lichen consists of an alga living symbiotically with a fungus. The latter cannot manufacture food but absorbs and retains water. Examine (*hp*) a cross section through the body of a lichen, distinguishing between the alga and the fungus. State exactly how each of the two lichen components derives benefit from its association with the other.

Sketch several kinds of lichens and make a drawing of a portion of a lichen body as seen in cross section.

Nitrogen-fixing Bacteria.—Examine root systems of beans or of some other leguminous plant and note the numerous *tubercles* or *nodules*. These contain bacteria that have invaded the roots from the soil, and the roots have responded to their presence by undergoing local enlargement. The bacteria live as parasites upon the root cells but have the power of forming nitrogenous compounds directly from the free nitrogen of the air. How is the bean plant benefited by the presence of the bacteria? Why is this a case of symbiosis?

Sketch a portion of the root system showing the tubercles.

ORGANIC EVOLUTION

Exercise 96

VESTIGIAL STRUCTURES

The snail and slug belong to the same class of mollusks. The former has a well developed shell, but the latter has none. Observe specimens of the common garden slug and note the muscular patch on its back. This is the *mantle*, which in the snail is a thin fold of skin lining the shell. Embedded in the mantle of the slug is a small, hard, calcareous plate. What does this represent and what does its presence signify as to the ancestry of the slug?

Make a drawing of the slug.

In a specimen of a bluebottle fly or of a large housefly, note (*m*) the presence of but one pair of wings. This is a characteristic of all other true flies, gnats, and mosquitoes. Nearly all other insects have two pairs of wings. Observe in the fly, however, the occurrence of a pair of vestigial wings, called *balancers*, posterior to the functional wings. They are minute, white, knobbed structures covered by the bases of the large wings. What is their significance?

Make an enlarged drawing to show the vestigial hind wings in the bluebottle or housefly.

Examine a specimen of the human sacrum showing the row of coccygeal bones at the end of the spinal column. In some individuals they are more or less fused together. Draw, and explain the significance of the human coccyx.

Exercise 97

SIMILARITY IN EMBRYOS

Examine a specimen of a young human embryo (or of some other mammal), noting the head, the gill slits, the limb buds, and the tail. Compare with other mammalian embryos of approximately the same stage of development, noting points of resemblance and of difference between them. Between which two

different kinds of embryos do you find the greatest amount of similarity? Why? How do you explain the presence of gill slits and a two-chambered heart in the young embryos of vertebrates that never have functional gills during their lifetime? What meaning has the presence of a tail in the early human embryo?

Make an enlarged drawing of any one of the embryos studied.

Exercise 98

KINDS OF FOSSILS

Examine fossils representing *actual organic remains*, such as bones embedded in asphalt, insects in amber, or some other type of material. How have they been preserved? To what extent has the original organic matter been altered? Also study *petrified fossils*. Of what kind of material are they composed? Because of the replacement, molecule for molecule, of the organic matter by inorganic, the original structure is accurately preserved. The commonest fossils are *natural molds and casts*. They represent impressions in rock of the original organisms. Examine whatever material is available.

Briefly describe the general appearance of each of the three types of fossils studied, and explain how each was formed.

Exercise 99

EVOLUTION OF THE HORSE

Through a study of fossils, the evolution of the horse has been traced through a series of progressive stages, represented by the following forms: *Eohippus*, *Mesohippus*, *Merychippus*, *Pliohippus*, and *Equus*. Study plaster casts of the skull and feet of these forms (the actual bones if available), and give the following facts about them, arranging your data in the form of a table:

Total length of fore and hind feet; number of functional toes present on each and their relative size; number of vestigial toes. Total length of skull, relative size of lower jaw, position of eye orbits; character of the teeth.

Consult your textbook and add to the table the name of the geologic epoch during which each horse lived, and the height of the animal.

Exercise 100

EVOLUTION OF MAN

Study the skull of the chimpanzee, a modern anthropoid ape closely related to man but not in the same direct line of ancestry. Compare it with a skull of modern man. If plaster casts are available of skulls (or portions thereof) of the Java ape man (*Pithecanthropus*), the Neanderthal man, the Piltdown man, and the Cro-Magnon man, include them in your comparative study. Give the following facts concerning each specimen, presenting them in tabular form:

Total number of teeth; relative size of canine teeth; relative size of frontal portion of cranium; development of eye arches; slope of forehead; character of lower jaw.